New Trends in Soil Biology

Proceedings of the VIII. Intl Colloquium of Soil Zoology. Louvain-la-Neuve (Belgium). August 30 - September 2, 1982. Edited by Ph. LEBRUN, H.M. ANDRÉ, A. DE MEDTS, C. GRÉGOIRE-WIBO and G. WAUTHY. Imprimeur DIEU-BRICHART, Ottignies-Louvain-la-Neuve (1983).

Intestinal mucus production by two species of tropical earthworms : Millsonia lamtoiana *(Megascolecidae) and* Ponthoscolex corethrurus *(Glossoscolecidae)*

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Summary :

Intestinal mucus contents is on average 6.6 ± 1.6 % in the anterior part of the gut of *M. lamtoiana* and 15.9 ± 2.5 % in *P. corethrurus*.

The concentration decreases significantly in the posterior part of the gut (2.8 ± 0.8 % in *M. lamtoiana* and 12.7 ± 3.6 % in *P. corethrurus*) since this watersoluble matter is reabsorbed, it drops down to 0.43 % in the fresh casts of *P. corethrurus*, a value still 3.6 times greater than in the ingested soil.

Annual production of intestinal mucus would raise up to 50 t/ha for the *P. corethrurus* population of a Mexican tropical pasture *i.e.* 20 % of the total organic matter contents of the soil.

Key words : tropical earthworms, intestinal mucus, digestion.

I. Introduction

The nature, role and production of earthworm intestinal mucus is still little known. VAN GANSEN (1962) showed the presence of mucus secretory cells in the crop and foregut of *Eisenia foetida*. RANGEL (in prep.) using PAS, a specific stain for neutral mucopolysaccharides, has demonstrated that these molecules are produced in the foregut of *Ponthoscolex corethrurus*. This mucus can be directly observed in the gut contents of most earthworms and it may be so abundant that it becomes necessary to remove it when studying the feeding habits of detritivore species (BOUCHÉ & KRETZSCHMAR, 1974; KANYONYO, 1981).

Although the main function of the mucus is assumed to be lubrication, it is hard to believe that the role of such an energy-rich and digestible substrate would be restricted only to that. In this work, we measured the amount of intestinal mucus mixed with the gut contents of two species of tropical earthworms : *Millsonia lamtoiana*, an anecic species from the savannas of the Ivory Coast, which weighs up to 32 g fresh wt. in the adult stage, and *P. corethrurus*, a mesohumic endogeic species much smaller in size (1 g f. wt. in the adult stage), which is very common in the tropical areas of Mexico as well as in many other humid tropical areas of America, Asia and Africa.

Annual mucus production of field populations of these earthworm species was extrapolated from calculated data of soil and litter ingestion.

II. Methods

BOUCHÉ & KRETZSCHMAR (1974) removed the mucus from the gut contents by extraction with a weak ammonia solution. This method was first applied to *M. lamtoiana*. Since extraction with water at 60° C gave the same result it was used with *P. corethrurus*.

The worm was first fixed in absolute alcohol, then cut in two or three sections corresponding to the anterior, middle and posterior gut. The gut was opened and its contents removed with a little spatula or by a water jet.

The extracted soil was dried in an oven at 60° C and weighed. It was then mixed with 10 volumes of hot water or solution of ammonia and filtered. The aqueous extract was dried, weighed and calcinated in order to calculate the proportion of organic matter contained. This water soluble fraction was assumed to consist mainly of the intestinal mucus.

III. Results

The sample of *M. lamtoiana* was made of 23 individuals collected throughout the year 1972 during a quantitative study and preserved in absolute alcohol. Ten were immature and thirteen were adults. The sample of *P. corethrurus* comprised 35 recently collected adults from tropical pastures in the state of Veracruz (Mexico).

TABLE I

Proportion of watersoluble matter contained in the different parts of the gut of *M. lamtoiana*.

Part of the gut	Number of individuals	% watersoluble organic matter	95 % confidence limits
Crop-gizzard	16	3.78	2.11-5.45
First third	23	6.55	4.94-8.16
Second third	22	4.17	2.91-5.43
Posterior third	19	2.76	1.96-3.56

The gut contents of young *M. lamtoiana* whose weight varies from 0.2 to 0.5 dry weight and of the adults whose weight reaches 4 g could be separated into four successive parts (Table I). The soil taken from the crop and the two gizzards already contains 3.8 ± 1.7 % water soluble organic matter, a value clearly greater than the one measured in the control soil (0.2 to 0.5 %; LAVELLE *et al.*, 1983). The concentration rises up to 6.55 % ± 1.6 % in the anterior part of the gut and then decreases significantly (U-test) towards the middle (4.17 ± 1.3 %) and posterior part (2.76 ± 0.8 %). The values measured in the anterior part are greater in the young worms (7.7 %) that in the adults (5.4 %) ($\alpha_o < 15$ %).

The concentration measured in *P. corethrurus* is greater : it reached 15.9 ± 2.5 % in the anterior part and decreased significantly (U-test) in the posterior part (12.7 ± 3.8 %); these values are much greater than the ones measured in the control soil (0.12 ± 0.04 %) and casts, whether they are freshly produced (0.43 %) or already dry and deposited 36 hours previously (0.28 ± 0.03 %).

IV. Annual intestinal mucus production by natural populations

The population of *M. lamtoiana* observed in 1972 at Lamto had a mean density of 14300/ha and a mean biomass of 84 kg. They ingested a total of 37 t dry soil and 540 kg

TABLE II

Proportion of watersoluble organic matter contained in the anterior and posterior half of the gut of *P. corethrurus*, fresh and dry casts and the non-ingested soil.

	Number of individuals	**% watersoluble organic matter**	**95 % confidence limits**
Anterior half	35	15.90	13.4-18.4
Posterior half	34	12.80	9.1-16.3
Control soil	10	0.12	0.08-0.16
Fresh casts	1	0.43	
Dry casts	10	0.28	0.23-0.33

litter (LAVELLE, 1978). Assuming that 6.55 % of intestinal mucus was added to this soil, the annual production can be estimated to be 239 kg/ha/year.

The population of *P. corethrurus* studied in 1981-82 in the area of Laguna Verde (Veracruz, Mexico) had a mean density of 147000 individuals weighing 300 kg fresh weight/ha (LAVELLE, 1982). Extrapolating some culture results (LAVELLE & CRUZ, 1983), we estimate that 300 t/ha (200 to 400) of soil passed anually through the guts of these earthworms. Thus, the annual mucus production would be about 50 t/ha. As this mucus contains 0.63 % nitrogen, the total amount of nitrogen annually integrated into it would be of about 315 kg/ha.

V. Discussion

The production of mucopolysaccharides in the gut of earthworms in ecosystems where the latter are numerous, seems to represent an important loop of the carbon and nitrogen cycles in the soil. In the tropical pastures of Laguna Verde, 50 t/ha of intestinal mucus are produced by the population of *P. corethrurus*. This is equivalent to 20 % of the total organic matter of the soil (estimated to 242 t/ha) and 2.2 times the amount of organic matter contained in the ingested soil (*i.e.* 23 t/ha/year). Most of the watersoluble elements resulting from the catabolism of this mucus and the ingested organic matter are reabsorbed in the posterior gut. As a matter of fact, watersoluble contents of fresh casts is only 0.43 % which is still 3.6 times greater that in the non-ingested soil and would represent an annual excretion of about 1 t/ha.

Numerous fields of investigations could be developed on the basis of our results. It seems necessary first to analyse in details the extracted watersoluble fraction, assumed in this work to be the intestinal mucus. It is evident that this fraction includes watersoluble elements from the soil, albeit in negligible quantities, mucopolysaccharides produced by the worm and the products of their catabolism and, at last, products of the digestion of insoluble organic matter from the ingested soil.

The watersoluble content is maximum in the foregut, where the digestion begins. This would suggest that most of these watersoluble elements are mucopolysaccharides and products of their metabolism. In the medium and hindgut would progressively substitute hydrosoluble molecules produced by the digestion process. At the same time, molecules coming from the degradation of mucopolysaccharides would be consumed by the intestinal microflora or progressively reabsorbed, and thus disappear. A detailed study of these processes would be rewarding.

In addition, it is of interest to explain the ecological significance of this digestion process. Geophagous earthworms feed on soil organic matter. Only a low fraction of it is watersoluble and directly assimilable. In order to digest more complex molecules, two ways are possible; one is digestion by enzymes, the other is microbial symbiosis. Both ways would exist in earthworms, but the second one seems to be the more important, especially in geophagous earthworms. Few studies of the enzymatic equipment of earthworms have been published and it is often difficult to tell if these enzymes have really been produced by the worm and not by its intestinal microflora. They generally show that the digestion ability of the earthworm is low. On the contrary, studies comparing the microbial activity of castings to that of non-ingested soil show a great variety of possible modifications in the gut microflora. Microbial activity can be activated or inhibited, but the response may be different for the diverse functional groups of micro organisms (see review in EDWARDS & LOFTY, 1972).

Thus, intestinal mucus could play the role of a chemical mediator in such processes. Its extreme instability and high energetic value would permit the rapid growth of a symbiotic microflora which would feed on this substrate; next, the microflora would feed on the more complex molecules from the soil organic matter and digest them, to the benefit of the worm. Hence, this great mucus production would transform the foregut into a highly active medium in which it would be possible to digest the organic matter in the time that the gut transit lasts, *i.e.* two to four hours at the temperatures of 20 to 25° C in these soils.

The amount of mucus produced is greater in the geophagous species *P. corethrurus* than in the anecic *M. lamtoiana*; it is also greater in the young *M. lamtoiana* than in the adults. Variations in production together with possible variations in the chemical composition of mucus thus could explain, at least partly, the subtle interaction mechanisms among soil organic matter, microflora and earthworms, as evidenced in the case of the mesohumic endogeic species *M. anomala* (LAVELLE *et al.*, 1983).

Acknowledgements

Part of this work has been supported by C.N.R.S. (L.A. n° 258). We are greatly indebted to Drs.A. CARTER and S. VINOGRADOV for the revision of the text and to Dr. R. LAFONT for his help in some analysis.

References

BOUCHÉ (M.) & KRETZSCHMAR (A.), 1974. — La fonction écologique des Lombriciens. II — Recherches méthodologiques pour l'analyse du sol ingéré. — *Rev. Ecol. Biol. Sol.*, **11** : 127-139.

EDWARDS (C.A.) & LOFTY (J.R.), 1972. — *Biology of Earthworms.* — Chapman and Hall Ltd., London, 283 p.

KANYONYO (J.), 1981. — Étude préliminaire du régime alimentaire du Ver de terre détritivore *Millsonia lamtoiana* (Acanthodrilidae-Oligochètes) dans la savane de Lamto (Côte d'Ivoire). — Rapport DEA, Université PARIS VI, 46 p.

LAVELLE (P.) & CRUZ (I.), 1983. — Soil ingestion, growth and fecundity of the earthworm *Ponthoscolex corethrurus* (Glossoscolecidae) in relation to soil water content. (en préparation).

LAVELLE (P.), ZAIDI (Z.) & SCHAEFER (R.), 1983. — Interactions between earthworms, soil organic matter and microflora in an African savanna soil. *In* Ph. LEBRUN, H.M. ANDRÉ, A. DEMEDTS, C. GRÉGOIRE-WIBO & G. WAUTHY (Eds.), *New Trends in Soil Biology.* — Imprimeur Dieu-Brichart, Ottignies-Louvain-la-Neuve : 253-259.

VAN GANSEN (P.S.), 1962. — Structures and functions of the digestive tract of the lumbricid *Eisenia foetida* Savingy. — *Journal of Microscopy*, **1** : 363.

Discussion

LEE, K.E. : According to your data, the total amount of organic matter ingested by earthworms is 23 t/ha/year and the total amount produced by the earthworms in mucus is *ca* 50 t/ha/year. An additional 27 t/ha/year of organic matter has appeared in the ecosystem, if these figures are correct. Can you please explain this apparent anomaly ?

LAVELLE, P. : The 50 t of intestinal « mucus » are excreted in the foregut; the products of their metabolism and the products of the digestion of the ingested organic matter (23 t/ha) will be *reabsorbed* in the hindgut and recycled within the worm. Therefore, in the casts we find a low hydrosoluble organic content (0.43 % versus 0.12 % in the surrounding soil) and a lower total C-content than in the surrounding soil.

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